APPENDIX D

INSERTED SECTION 2 – SPECIFICATION – CLEAN VERSION

The supernatant from a hybridoma designated as 5G1.1 tested positive by ELISA and substantially reduced the cell-lysing ability of complement present in normal human blood in the chicken erythrocyte assay. Further analyses revealed that the 5G1.1 antibody has two surprising properties: 1) it reduces the cell-lysing ability of complement present in normal blood so efficiently that, even when present at roughly one-half the molar concentration of human C5 in the hemolytic assay, it can almost completely neutralize serum hemolytic activity; and 2) it binds to both the alpha and beta chains of the human C5 protein.

The surprising and unanticipated ability of the monoclonal antibody produced by hybridoma 5G1.1 (the 5G1.1 mAb) to bind to both the alpha and beta chains of the human C5 protein was revealed when immunoblot analysis was undertaken to further characterize the 5G1.1 mAb. Human C5 (Quidel Corporation, San Diego, Calif., Catalog No. A403) was subjected to polyacrylamide gel electrophoresis under reducing conditions, transferred to a nitrocellulose membrane, and probed with the 5G1.1 mAb as a purified IgG preparation. Two bands were immunoreactive with

INS 48 the 5G1.1 mAb at apparent molecular weights corresponding to those of the alpha and beta chains of the human C5 protein.